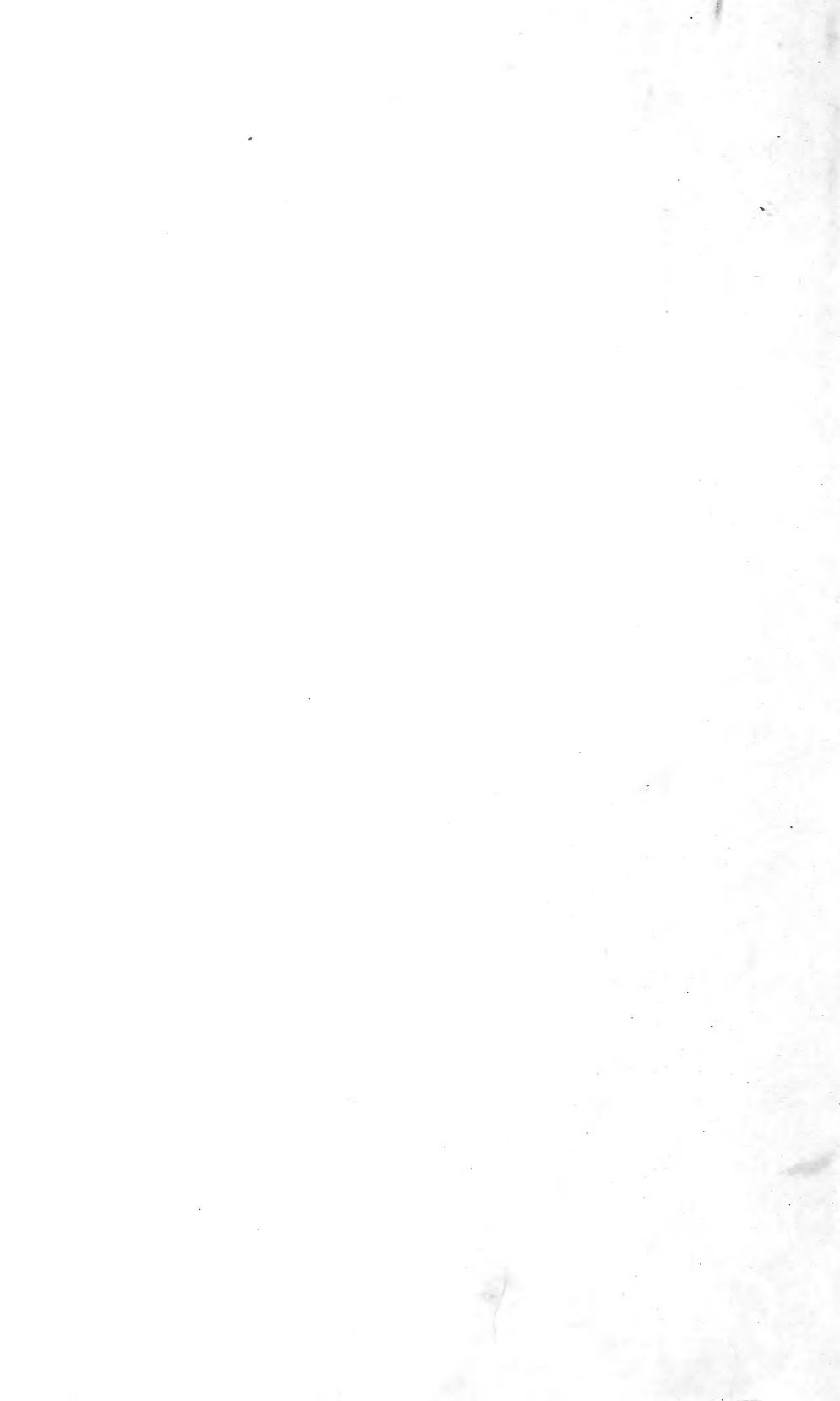


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BULLETIN OF THE U.S. DEPARTMENT OF AGRICULTURE

No. 148



Contribution from the Bureau of Animal Industry, A. D. Melvin, Chief.
March 22, 1915.

THE USE OF *BACILLUS BULGARICUS* IN STARTERS FOR MAKING SWISS OR EMMENTAL CHEESE.

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INTRODUCTION.

The Swiss-cheese industry was introduced and is still carried on in the United States by settlers from Switzerland who were cheesemakers in their native land. Not many of them remain in the business in this country for any length of time, however, mainly because of the long hours of labor necessary under the present system of making cheese twice a day. But this system was inevitable until a sufficient knowledge of fundamental principles could be obtained so that the method of making the cheese could be altered without injuring the quality of the product. As an art Swiss cheesemaking is very highly developed, but it is based on empirical methods. Few scientific principles have been found that are a help to the cheesemakers even in Switzerland, where the industry has been well established for a long time.

Although some very fine cheese of the Swiss or Emmental type has been made in the United States, the quality has not averaged so high as that of the foreign-made cheese. The feed, pastures, climate, topography, and other conditions, so different from those in Switzerland, where the present system of Swiss-cheese manufacture was developed, naturally call for changes which could not be made in the absence of a knowledge of the causes which underlie the processes of cheesemaking. Another contributory cause of low-grade American-made cheese has been the inadaptability of many of the cheese factories; their fitness for cheesemaking has sometimes been sacrificed to cheap construction. So many difficulties have been experienced that cheesemakers were led to believe that it was impossible to make a good Swiss cheese except in a few localities. Some believed, in fact, that there was no place in the United States where

NOTE.—This bulletin reports experimental work showing how to control undesirable fermentations and thus to provide a remedy for the most serious troubles which occur in making Swiss or Emmental cheese. It is of interest chiefly to manufacturers of that type of cheese.

the product would equal in quality that made in Europe, since the same methods were used on both sides of the Atlantic.

The most serious trouble of the cheesemaker occurred during the cold months, which led to the practice of making cheaper varieties of cheese in the spring and fall and closing down the factory for four months in the winter. This, of course, is a considerable handicap to the industry, and would not be necessary if there were sufficient information concerning the origin of and remedy for the faults in manufacture. These unsatisfactory conditions led to the investigations reported in this bulletin, since it was believed that the present faulty methods might be corrected, provided the real causes of cheese defects were discovered.

In the absence of exact knowledge it was natural that erroneous theories should become prevalent in regard to the feeding of the cows, the care of the milk, and the handling of the cheese; but as they were based on practical experience it has not been found advisable to set them aside without investigation. Apparently very unimportant changes made in handling the cheese were found to result in great changes in the quality of the finished product, and although changes in methods are necessary in order to produce the best quality of cheese, it is unwise to advise the cheesemaker to change his methods without substantial proof of the value of the change.

The main trouble in making Swiss cheese is known to be caused by the development of undesirable types of microorganisms, some of which produce abnormal gas, causing what is known as "nissler" or "pressler" cheese. These undesirable organisms in Swiss cheese cause a lack of uniformity in the formation of the eyes. In some cases no eyes whatever are developed; this trouble is probably due to the absence of certain desirable types. At the beginning of this work it was thought that these faults might be overcome by the proper use of starters, which have become general in buttermaking, and their value has been frequently demonstrated. They have also been used to some extent in the making of Cheddar cheese. Unconsciously the makers of Swiss cheese have used starters with the rennet, a practice which has at times been of great value. But while the rennet starter has been the cause of much help, it has also caused trouble when the helpful species of bacteria usually present have for some reason been weakened. The full benefit of the starter was not obtained, in any event, since less than one-fourth of 1 per cent of rennet was used.

THE SIGNIFICANCE OF *BACILLUS BULGARICUS* IN MAKING SWISS CHEESE.

In selecting a starter for making Swiss cheese it is at once apparent that certain characteristics are desirable to make its use possible with the method of manufacture employed. The curd for Swiss

cheese is cooked at a comparatively high temperature, 126° to 136° F., and is cooled very gradually while the cheese is in press. This treatment checks temporarily the growth of most species of bacteria, including the lactic-acid bacteria, which are used for starters in the making of butter and Cheddar cheese.

The *Bacillus bulgaricus* group of bacteria has the qualifications which apparently fit in with the manufacturing process of Swiss cheese. Investigators have found a wide variation in the temperatures at which different varieties will grow and in the amount and rapidity of acid formation. The presence of this group of bacteria in the rennet preparations was first recognized by Freudenreich and Jensen,¹ who studied it and named it *Bacillus casei* ϵ . They came to the conclusion that it was largely responsible for the normal ripening of the cheese, but Jensen apparently receded from this position a few years afterwards, though he still advocated the use of *B. casei* ϵ in the preparation of rennet for the purpose of suppressing the growth of undesirable bacteria in the rennet solution. He has been supported in this by many of the European authorities, and pure cultures of *B. bulgaricus* have been furnished extensively to makers of Swiss cheese for this purpose.

Peter and Held,² in discussing the sources of infection causing troubles with Swiss cheese and the influence of the rennet solution on the cheese, suggested the possibility of the cultures in the rennet suppressing undesirable gas-forming types of bacteria in the cheese.

Gratz,³ in some laboratory tests, found that a culture of *Bacillus bulgaricus* inhibited the growth of bacteria of the coli-aerogenes group in milk held at a temperature of 40° C. (104° F.).

Burri,⁴ in discussing the relative merits of commercial acid and pure cultures of *Bacillus bulgaricus* in making up the whey rennet solution, points out that *B. bulgaricus* suppressed the growth of gas-forming bacteria in the whey rennet.

Thöni⁵ showed the influence of *B. bulgaricus* in making good rennet. He reported some experiments in which rennet containing *B. bulgaricus* made good cheese, while cheese made with the natural rennet without this bacillus was gassy, evidently because the gas-producing bacteria made a very heavy growth in the rennet.

¹ Freudenreich, Edward von, and Jensen, Orla. Die Bedeutung der Milchsäurefermente für die Bildung von Eiweisszersetzungsprodukten in Emmenthalerkäsen, nebst einigen Bemerkungen über die Reifungsvorgänge. Landwirtschaftliches Jahrbuch der Schweiz, vol. 13, p. 169-197. Bern, 1899.

² Peter, A., and Held, J. Praktische Anleitung zur Fabrikation und Behandlung des Emmenthalerkäses. Second edition. Bern, 1910.

³ Gratz, Otto. Studien über die Antibiose zwischen Bacterium casei ϵ und den Bakterien der Coli-Aerogenes-Gruppe. Zeitschrift für Gärungsphysiologie allgemeine, landwirtschaftliche und technische Mykologie, vol. 1, no. 3, p. 256-281. Berlin, June, 1912.

⁴ Burri, Robert. Reinkulturen oder Säuremischung beim Labansatz? Molkerei Zeitung, vol. 22, no. 33, p. 387-389. Berlin, 1912.

⁵ Thöni, Johannes. Bakteriologische studien über Labmügen und Lab. Ein Beitrag zur Kenntnis der Bereitung des Käsereiblases. Landwirtschaftliches Jahrbuch der Schweiz, vol. 20, p. 181-242. Bern, 1906.

Jensen¹ advised the use of a streptococcus in connection with *B. bulgaricus* as a starter for suppressing undesirable bacterial growths in the cheese, but offered no proof of the efficiency of this combination of cultures.

Though it is generally believed that it is the lactic acid produced by different bacteria that gives different varieties their value in preventing the growth of undesirable forms of germ life, this assumption is seriously questioned by some bacteriologists, who think it possible that the formation of lactic acid is incidental and is not the active inhibiting principle.

White and Avery² point out that cultures of *B. bulgaricus* grow at relatively high temperatures, forming acid as high as 50° C. (122° F.). They also show that relatively high percentages of acid are formed in milk, reaching as high as 3.1 per cent.

Hastings and Hammer³ give 4.09 per cent as the maximum amount of acid found in milk. They find that *B. bulgaricus* is distributed very widely and generally in dairy products of all kinds. Mention is made especially of its presence in the milk and whey at Swiss-cheese factories.

Heinemann and Hefferan⁴ also noted the general distribution of *B. bulgaricus*, its high growing temperature and its ability to form acid in milk.

The authors quoted found a very great difference in the maximum amount and the rapidity of acid formation of different cultures. Cultures also lose their ability to form acid to a great extent when carried under laboratory conditions. The growth of *Mycoderma* on the surface of the whey starter greatly facilitates the growth of the *B. bulgaricus* culture used. Thöni⁵ in some tests with the *Mycoderma* found that while the whey culture of *B. bulgaricus* without the *Mycoderma* showed at the end of 24 hours 7,000,000 and 18,000,000 bacteria per cubic centimeter, with the *Mycoderma* the numbers were 136,000,000 and 200,000,000, respectively, and the increase of acid with the *Mycoderma* was more than one-half.

In our own work we have found that *B. bulgaricus* can form as high as 2 per cent acid in whey, and we found that with the culture iso-

¹ Jensen, Orla. Ueber die im Emmentalerkäse stattfindende Milchsäuregärung. Milch-wirtschaftliches Zentralblatt, vol. 2, no. 9, p. 393-414. Leipzig, Sept., 1906.

² White, Benjamin, and Avery, Oswald T. Observations on certain lactic-acid bacteria of the so-called bulgaricus type. Centralblatt für Bakteriologie, Parasitenkunde und Infektionskrankheiten, Abteilung 2, vol. 25, no. 5/9, p. 161-178. Jena, Nov. 30, 1909.

³ Hastings, Edwin George, and Hammer, B. W. The occurrence and distribution of organisms similar to *B. bulgaricus* of yogurt. Centralblatt für Bakteriologie, Parasitenkunde und Infektionskrankheiten, Abteilung 2, vol. 25, no. 14/18, p. 419-426. Jena, Dec. 22, 1909.

⁴ Heinemann, Paul Gustav, and Hefferan, Mary. A study of *Bacillus bulgaricus*. Journal of Infectious Diseases, vol. 6, no. 3, pp. 304-318. Chicago, June 12, 1909.

⁵ Loc. cit.

lated at Albert Lea, Minn., the most favorable temperature was 49° C. in making Swiss cheese began in the winter of 1910-11 at Albert Lea,

EXPERIMENTS WITH BACILLUS BULGARICUS STARTERS.

Our experiments with cultures of *Bacillus bulgaricus* as a starter in making Swiss cheese began in the winter of 1910-11 at Albert Lea, Minn. The work was continued at State College, Pa., and finally completed in the laboratories at Washington. The milk delivered for cheesemaking at Albert Lea was of very poor quality, being very gassy, and it was impossible to secure any that was not badly infected. A long series of experiments in pasteurization had not proved entirely successful, and experiments to discover a bacterial culture that would prove efficient in suppressing gas-forming bacteria were begun. A number of different cultures of *B. bulgaricus* were used under varying conditions and at all seasons of the year.

In Table 1 are given the results obtained with *B. bulgaricus* cultures that proved efficient in suppressing gas-forming types of bacteria when used in what would probably be considered as reasonable amounts of starter; that is, where the starter was less than 2 per cent of the total amount of milk used. The cultures used were obtained from different sources. Culture 39a was very active and was the only culture of *B. bulgaricus* used at Albert Lea, Minn.¹ Cultures I S and 44H were isolated in the Washington laboratories.

In the experiments recorded in Tables 1 and 2 all the milk was first put into one kettle, where it was thoroughly stirred and then divided. As the kettle and all other apparatus used were thoroughly cleaned before using, identical conditions in both lots of milk were insured.

Probably every lot of milk used in these experiments was as badly contaminated with gas-forming bacteria as the mixed milk would be in any commercial Swiss-cheese factory on any day of the year. Nevertheless from this milk, by the use of these starters, we were enabled to make a perfectly sound cheese that did not develop into a "nissler" or "pressler."

The milk used at the Washington laboratories came from the herd owned by the Dairy Division and was almost free from the faults common to ordinary factory milk. There may have been no occasion for using a starter with this milk to suppress gas-forming bacteria. At this time culture 39a, by being carried in the laboratory, had lost much of its power to form acid, but was still active enough to retain its efficiency in suppressing undesirable gas formers. A

¹ This culture was isolated by Mr. B. J. Davis, of this laboratory, who was in search of a strain of high acid-producing bacteria for pure cultures in buttermaking. This culture produced 3 per cent of acid in milk, but only 2 per cent in whey.

few additional tests were made with *Bacillus bulgaricus* starter, using fresh cow manure for contamination, and with cheese 31, Table 1, enough manure was added to the milk to give it a slight color. The result of this test, in which culture W was used, is seen in Plate I, which illustrates the efficiency of this starter on gas-producing bacteria. This test was perhaps no more severe than many others that were made. The two cheeses were cut 24 hours after making from the same lot of badly contaminated milk. The upper cheese shows the effect of the starter; the lower cheese, made without starter, is badly "nissler." This experiment was not made to determine whether or not contaminated milk would make good cheese, but to test the effect of the starter on milk heavily inoculated with gas-producing bacteria. The cheese, as was to be expected, was not a normal cheese; it had a strong, bitter taste, but abnormal gas formation was entirely suppressed.

TABLE 1.—Showing suppression of gas formation in Swiss cheese by the use of *Bacillus bulgaricus* cultures in normal amounts.

Cheese No.	Culture No.	Starter.		Condition of cheese.	
		Amount.	Acidity.	Starter.	No starter.
		Per cent.	Per cent.		
1	39 a	1	1	No gas.....	
2	39 a	1	1.2do.....	
3	39 a	1	.9	Slightly gassy.....	Very gassy.
4	39 a	1	1.1	Edge gassy.....	Do.
5	39 a	1	1.2	No gas.....	Do.
6	39 a	1	1.1do.....	Do.
7	39 a	1	1.4do.....	Do.
8	39 a	1	1.0do.....	Do.
10	39 a	1.5	.5do.....	Gassy.
11	39 a	1.5	.5do.....	Do.
12	39 a	1.5	.5do.....	Very gassy.
13	39 a	.66	.8	Slightly gassy.....	Do.
15	I S	1.5	.5	Edge gassy.....	Gassy.
16	I S	1.5	.5do.....	Very gassy.
17	I S	1.5	.6do.....	Do.
18	I S	1	.6	No gas.....	Gassy.
19	I S	1	.5	Slightly gassy.....	Very gassy.
20	44 H	1	.5	No gas.....	Gassy.
21	44 H	1	.4do.....	Slightly gassy.
22	44 H	1	.6do.....	Do.
23	44 H	1	.5do.....	Gassy.
24	44 H	1	.3do.....	Slightly gassy.
25	44 H	1.5	.5do.....	Very gassy.
26	44 i	1.5	.5	Edge gassy.....	Gassy.
27	44 i	1.5	.5do.....	Do.
28	44 i	.75	.4	Slightly gassy.....	Very gassy.
29	44 i	1.5	.5	No gas.....	Gassy.
30	44 i	1.5	.4	Edge gassy.....	Do.
31	W	2.0	.1	No gas.....	Very gassy.

Attention is called to cheeses 4, 15, 16, and 17, Table 1, where the outside surface or "edge" of the cheese to perhaps a half inch or 1 inch was badly gassy, while the center was entirely free from gas. The outside portion becomes cooled, allowing the gas-producing bacteria to damage this part of the cheese, while the inside portion remains at a temperature more favorable for the growth of *B. bulgaricus*. This illustrates excellently the fact that the long-continued

high temperature of cooking and pressing the curd is necessary to obtain the best results with *Bacillus bulgaricus*.

Table 2 gives the results with starters made from a *B. bulgaricus* culture that did not prove efficient unless used in comparatively large quantities, as much as 4 per cent of starter made with culture B being required thoroughly to suppress gas formers. Culture B was a common type of *B. bulgaricus*, causing stringiness; that is, stringy or slimy milk and whey. This was the only culture of *B. bulgaricus* used that gave questionable results.

TABLE 2.—*Effect of Bacillus bulgaricus starter (Culture B) on suppression of gas formation in Swiss cheese.*

Cheese No.	Culture No.	Starter.		Condition of cheese.	
		Amount.	Acidity.	Starter.	No starter.
		<i>Per cent.</i>	<i>Per cent.</i>		
1	B	1	0.3	No gas.....	Very gassy.
2	B	.66	1.2	Very gassy.....	Do.
3	B	.66	1.4	Gassy.....	Do.
4	B	.66	1.5	Very gassy.....	Do.
5	B	.5	2.4	do.....	Do.
6	B	1	.7	Slightly gassy.....	Do.
7	B	3	.8	Edge gassy.....	Do.
8	B	4	.9	No gas.....	Do.

More than 100 cheeses were made from milk known to be badly contaminated in which the starters showed positive results in securing good cheese. Only those cases in which companion cheeses were made from the same milk, one with and one without the starter, have been included in Tables 1 and 2.

Table 3 contains the results with starters made from types of bacteria which cause the normal souring of milk and which are usually found in commercially pure cultures advertised for use in creameries and Cheddar-cheese factories. A few trials were made with starters prepared from these cultures and another commercial culture, but with poor results.

TABLE 3.—*Results of using starters made from common lactic types of bacteria in making Swiss cheese.*

Cheese No.	Culture No.	Starter.		Condition of cheese.	
		Amount.	Acidity.	Starter.	No starter.
		<i>Per cent.</i>	<i>Per cent.</i>		
1	E	4	0.6	Very gassy.....	
2	E	4	.6	do.....	
3	E	2	.6	do.....	
4	E	2	.6	do.....	
5	E	4	.6	do.....	
6	S M	1	.6	do.....	
7	S M	1	.6	do.....	
8	S M	1	.6	do.....	
9	S M	2	.6	do.....	
10	S M	4	.6	Pin holes.....	

Some experiments not recorded in these tables showed very definitely that the temperature of cooking the curd had a very material influence on the effectiveness of *Bacillus bulgaricus* starters. In a number of tests it was found that cheeses cooked at 125° F. or above were entirely free from gas. It appears probable that the higher cooking temperature served to check the growth of undesirable bacteria for a longer period or until the *Bacillus bulgaricus* secured a sufficient start to check their growth.

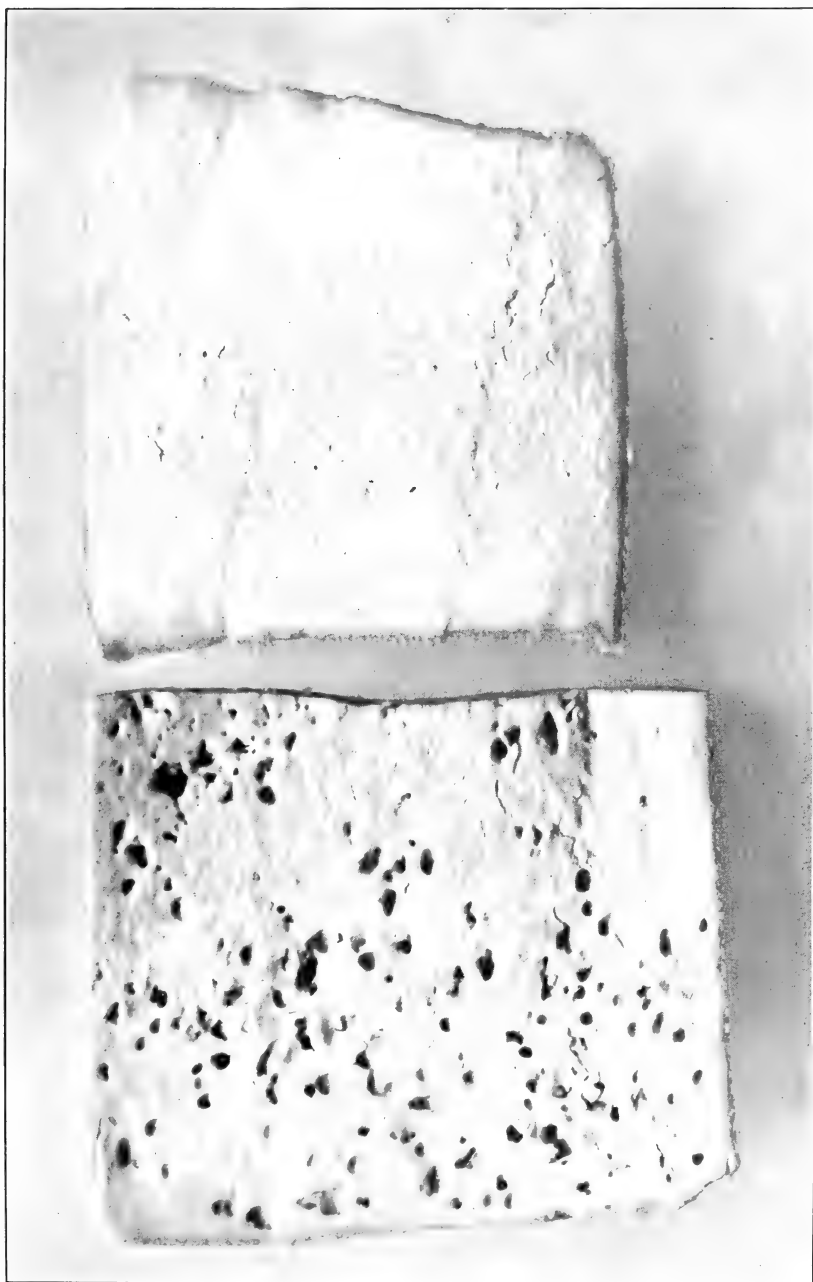
EXPERIMENTS WITH *BACILLUS BULGARICUS* STARTERS IN A COMMERCIAL FACTORY.

The results of our experiments led us to believe that we could demonstrate two points: First, that it was possible to overcome the faults of milk usually delivered to Swiss-cheese factories early in the spring; and, second, that it was possible to secure a normal eye growth with attending characteristic flavors by the use of cultures adapted for that purpose in cheese made early in the spring and in cheese made once a day instead of twice a day, as is the usual custom in summer. In order to study the use of *Bacillus bulgaricus* starters in a commercial factory, one of the authors spent two weeks, beginning the last week in April, 1913, in a factory where cheaper products (brick and Limburger cheese) were made early in the spring, or until the weather conditions became favorable for the making of good Swiss cheese. The value of these cultures should have been demonstrated earlier in the spring, but the factory did not receive milk enough to make one cheese a day until about May 1. The result of this work is compiled in Table 4.

TABLE 4.—Results obtained with *Bacillus bulgaricus* starters in suppressing gas formation in Swiss cheese under commercial conditions.

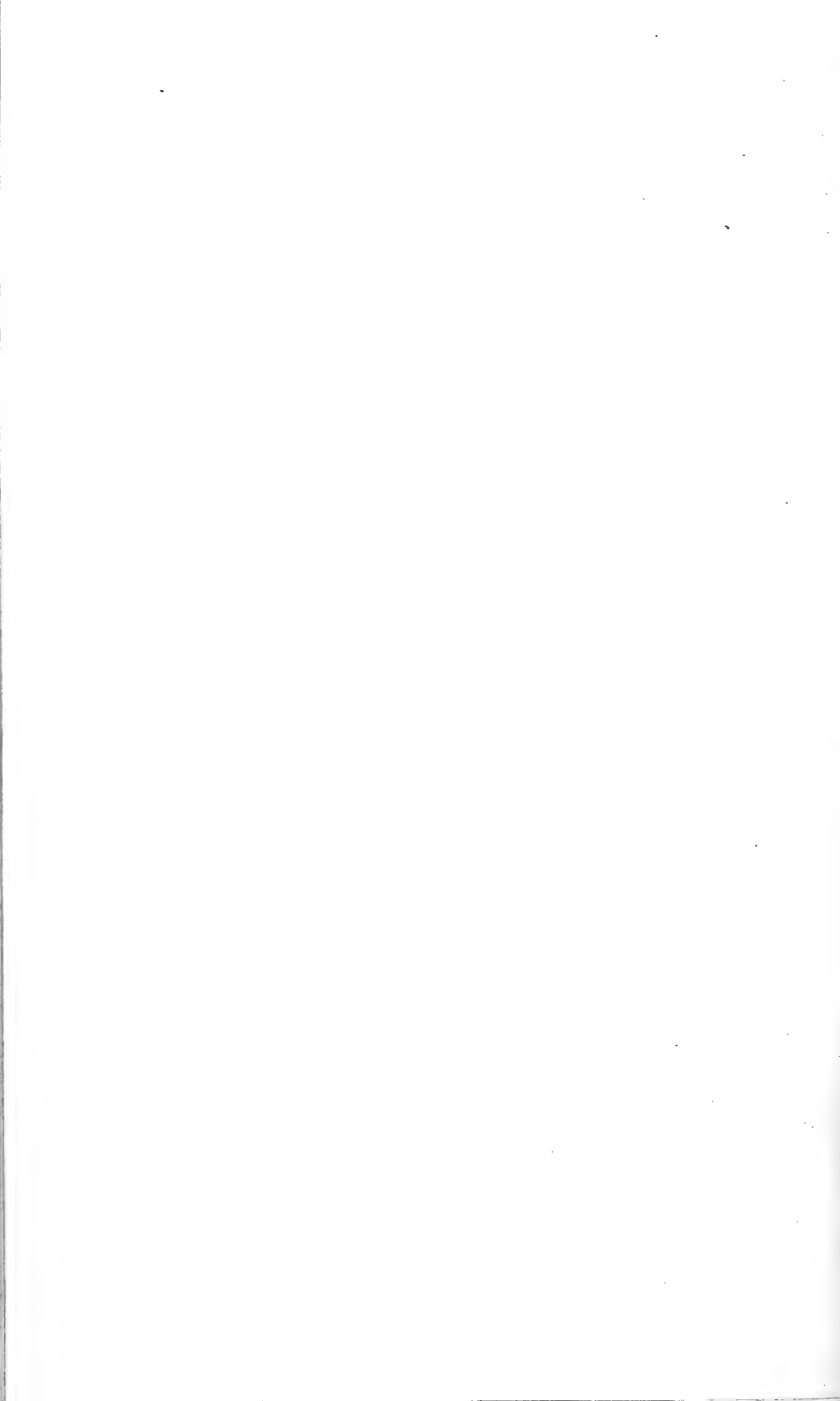
Date.	Cheese No.	Starter.		Condition of cheese.
		Amount.	Acidity.	
		Per cent.	Per cent.	
May 1.....	1	1	1.0	Nissler.
2.....	2	1.5	.7	Do.
3.....	3	1.5	.9	Do.
4.....	4	1.5	.9	Do.
5.....	5	1.5	.8	Do.
6.....	6	1	.9	Do.
7.....	7	1	.6	Do.
8.....	8	1.25	1.0	Good.
9.....	9	1.25	1.1	Do.
10.....	10	1	1.5	Do.
11.....	11	1.25	1.2	Do.
12.....	12	1.5	1.1	Do.
13.....	13	1.5	1.4	Do.
14.....	14	1.25	.9	Do.

The starters used were prepared from pure cultures of *Bacillus bulgaricus* from the laboratory of the Dairy Division at Washington. As the temperatures were rather low at this time it was necessary to



EFFECT OF USING *BACILLUS BULGARICUS* STARTERS TO CONTROL FERMENTATIONS IN MAKING SWISS CHEESE.

These cheeses were cut 24 hours after making from the same lot of badly contaminated milk; the upper was made with starter, the lower, without any starter, is badly "nissler."



provide some means of keeping the starters at a temperature high enough to insure the growth of the *B. bulgaricus*. A well-insulated box was made for this, somewhat on the order of a fireless cooker, which maintained temperatures very satisfactorily on nights when the outdoor temperature was below freezing. Considerable difficulty with yeasts was experienced, making frequent changes of cultures necessary, as noted on another page. The first six cheeses in Table 4 turned out "nissler," which is an indication of undesirable gas formation. In all the experiments except No. 14 the cheese was made once a day, using a mixture of morning and evening milks. The farmers made no pretense of cooling the milk, and as a very large can holding about 200 pounds of milk is used, the large bulk of milk cooled very slowly, even on frosty nights. The night milk with this treatment had developed so much acid, or such a growth of lactic-acid organisms, that in one instance the whey contained 0.19 per cent of acid at the time the curd was dipped, whereas it should have contained normally but 0.12 per cent. In all cases there was a marked development of acid. From the results shown in Table 3, where the sour-milk starter designated in the table as SM was used in a commercial factory, it would appear that a high development of acidity in the milk from the growth of ordinary lactic-acid formers tends to give a "nissler" cheese. Apparently the milk must be sweet when delivered or the *B. bulgaricus* starters do not suppress all undesirable gas formers.

When it was found to be impossible in the short time at our command to induce the farmers to cool their evening milk, they were asked to deliver twice a day, as is the custom in the summer months. The night milk was then cooled in a kettle by means of a coil through which cold water was pumped. But as the temperature of the cooled milk was not lower than about 68° F., it could not be said that the milk was unusually well cared for. Beginning with experiment No. 7, for which the night milk was cooled at the factory, we had milk sweet enough to work up in a normal manner, and the cheeses were all perfectly free from undesirable gas formation.

The milk delivered generally to the factory was not of the best quality. The brick cheese made before these experiments was badly gassy, and fermentation tests made with the milk of individual patrons at short intervals during the experiments showed that more than one-half of the patrons delivered gassy milk, some of the samples being very bad. It appeared that the starters were of great assistance in overcoming serious trouble with the gassy milk. Ordinarily this factory would not have begun to make Swiss cheese for a month later than it was made successfully in these experiments, though the higher price of Swiss cheese is a great inducement to make this variety rather than brick cheese.

It is worthy of note in this connection that the good cheeses of this series, with one exception, developed very large, though too many, eyes; in fact, the cheeses were hurried to cold storage to prevent cracking from an excessive growth of eyes. No other cheese made at the factory the same season had a sufficient eye development, and no factory is able to secure a satisfactory eye growth so early in the season as this work was done. The unusual development of eyes in the cheese made in this experiment was secured only by the use of the cultures. The culture in this case was obtained from one of our own winter-made cheeses that would rival any imported in eyes, texture, and flavor. It was ground up and incubated in whey for 24 hours. In these experiments unusually heavy cheese starters were used, which possibly accounts for too great a growth of eyes. The cheese made in this factory, after the experiments were completed and when the culture was not used, did not in any case show a satisfactory growth of eyes. Some cultures of *Bacillus bulgaricus* favor the growth of eyes more than others, but the use of starters for securing eyes in Swiss cheese will be discussed in a subsequent paper.

From the results of the experiments with *Bacillus bulgaricus* cultures it would appear that with their proper use in starters, Swiss cheese can be made in winter as well as in summer. It also seems practicable to make cheese once a day. However, *B. bulgaricus* cultures will not be a "cure-all" for any condition of milk which careless farmers may be able to bring about, but if the milk receives as good care as it receives when the best quality of cheese is made at the present time, and the evening milk is cooled at once after milking so it will not have developed acid when delivered to the factory, there should be no serious difficulty with the help of a good starter in making good Swiss cheese once a day and every day in the year.

AMOUNT OF STARTER TO USE.

We have found that 3 per cent of starter can be used without any indication of harmful results in the ripening cheese. Probably a greater amount of a weak culture or of a starter with a low acid content at the time of use could be employed. But 3 per cent of a strong culture with a high development of acid in the starter is apparently all that it is safe to use. More than 3 per cent of starter has in some instances apparently suppressed all tendency to form eyes, while the use of as much as 3 per cent has no apparent injurious effect on eyes, texture, or flavor. With a strong, active culture of *Bacillus bulgaricus*, 2 per cent of starter would undoubtedly prove more than sufficient to insure a perfectly solid cheese, excepting under the most extreme conditions of a poor milk supply. It is suggested that this amount be used, since it can be safely employed.

HOW TO SECURE CULTURES.

The problem of securing cultures of *Bacillus bulgaricus* for making starters will, of course, be an important one. It would be desirable to have a commercial source from which the active, pure cultures could be obtained, but it will probably be some time before cultures can be obtained in this way. In the meanwhile a number of sources are open. Cheesemakers have, in fact, been using *B. bulgaricus* starters unconsciously, which indicates that this bacterium is ordinarily present in the milk or whey of all factories. This can be verified by allowing a sample of whey to stand for about 48 hours at 100° F. If an active culture of *bulgaricus* is present the whey will become so sour that it can early be detected by smell. It will be best, however, for the cheesemaker to provide himself with an acidimeter outfit to test the strength of the culture. This apparatus is simple, easy to operate, and can be obtained for less than \$5. Its cost may be saved on one cheese by insuring a good starter, and it can be used to advantage to find new cultures of *bulgaricus*. Unfortunately, although *B. bulgaricus* seems to be present almost uniformly in the whey in Swiss-cheese factories, it sometimes becomes suppressed, and under these conditions the rennet putrefies and may cause serious trouble in the cheese.

The *B. bulgaricus* organism is especially likely to be absent entirely or lost in the early spring or late fall under present conditions. This condition has been noted by many European writers who have published discussions on the comparative merits of pure cultures of *B. bulgaricus* and commercial acid (which goes under the commercial name of casol) for insuring a whey rennet that will be free from undesirable gas formers. But there need be no trouble from this source when an active culture of *bulgaricus* is present and proper temperatures are used. When the temperature conditions are not favorable for the growth of *B. bulgaricus* other types of bacteria may crowd it out, or it will develop so slowly as to have no effect. This accounts in a large measure, probably, for the poor results with Swiss cheese in the colder seasons and for the fact that even in Switzerland great difficulty is experienced in making good cheese in winter.

A good culture of *B. bulgaricus* should be able to produce a maximum of at least 1.5 per cent of acid in whey when carried 48 hours at 100° F., with an inoculation of less than 1 per cent. In our experience it was not difficult to find *B. bulgaricus* cultures that gave an acidity in whey of over 1 per cent in 24 hours carried at 100° F. However, it has been found necessary in the laboratory to change occasionally, as the cultures apparently become weakened.

When cheesemakers either lose their cultures or find by the use of the acidimeter that their cultures have become weakened, a number

of ways are open to renew or secure more active ones. Securing samples of whey from factories has proved to be the most satisfactory way of renewing cultures. This would be an easy method for a cheesemaker to follow. The Dairy Division often has samples of whey sent from Wisconsin factories, and will send out a limited number of the most active bulgaricus cultures to those who are equipped to use them and will report the results of their trials. Another way would be to set individual samples of milk from patrons at the proper temperature (about 100° F.) for five days and select the one that developed the greatest acidity. Still another way is to grind up a piece of Swiss cheese in whey and carry at a temperature of 100° F. for five days. We have used this last plan on different occasions, but it is open to two objections. The *B. bulgaricus* may lose its activity by being carried in the cheese. Again, in preparing a starter in this way the bacteria responsible for the eyes in the cheese grow with the *B. bulgaricus*, and the result is a cheese with a decided tendency to form too many eyes. This can in a measure be overcome by allowing the acidity to develop to a high point, which, judging from some of our results, kills the bacteria which form the eyes.

In all the work reported here whey was used in preference to milk for making the starter. It does not develop so high an acidity, but it has two advantages which are very desirable in cheesemaking. A milk starter contains casein coagulated with acid and therefore contains a large part of the living bacteria which do not become thoroughly distributed throughout the milk. Whey has neither of these faults. It is possible also that whey would make a more satisfactory starter for other purposes, though it might not furnish all the qualities desirable in butter-making.

KEEPING THE CULTURE AND STARTERS.

Since *Bacillus bulgaricus* requires a temperature of about 100° F. for rapid growth, the proper conditions are not hard to obtain in summer, and have been supplied to cheesemakers as a rule in their method of carrying their rennet, where the warm whey containing the rennet is set above the fireplace or boiler, and as a result the bulgaricus grows rapidly and provides a good starter. At other seasons some extra means must be provided to insure the right temperature throughout the period required for the starter to attain a sufficiently high acidity. Perhaps as simple a method as any would be a fireless cooker or a well-insulated box of any kind. A square box insulated with granulated cork and with a receptacle for holding the whey which just fitted the inside of the box proved satisfactory for our work. If a fireless cooker is used the plates or

stones sometimes placed in them for maintaining heat must be at the proper temperature and would have to be used with the greatest care or the starter would be heated too high and sterilized. In any case it would be desirable to warm up the starter twice a day.

At the present time the cheesemakers take whey for the rennet direct from the kettles, usually before the curd is cooked. The starter could be taken in the same way, though it would probably be better to take out this whey after the curd is dipped. The temperature used for heating the curd has no harmful effect on the *B. bulgaricus* while it checks temporarily the growth of most other organisms, probably including yeasts. This is permissible for carrying starters, though some writers have advised sterilizing the whey, which should not be done unless a mother starter is used for reinoculating. A putrefied rennet would be the certain result. Some have advised the use of rennet extract if the cheese shows any signs of abnormal gas formation. This might help if the gassy cheese were due to the loss of the bulgaricus culture, but as a general rule it would do more harm than good, for if the bulgaricus were present the use of a greater quantity of the whey would provide a better remedy.

Cheesemakers would probably insure themselves against occasional trouble from undesirable fermentation if they would set the whey at the usual temperature 24 hours before adding the dried rennet. This would give the *B. bulgaricus* present a chance to get a good start ahead of any putrefactive bacteria which might be carried by the rennet.

TROUBLE FROM YEAST.

Usually it would be desirable to carry a mother starter, or culture, in a separate vessel, the mother starter being the name given to the small starter carried over from day to day for inoculating the main starter. It is necessary to carry this mother starter so that it will not become contaminated with yeast, which is apparently the only foe of the cheesemaker that *Bacillus bulgaricus* starters will not help to control. All other contaminations of the starter were held in complete subjection by the *B. bulgaricus*. At the factory where our experiments were made the contamination from yeasts was very serious; although all vessels used were sterilized and the whey starter was boiled, the yeast on one occasion spoiled the starter. The conditions in cheese factories seem to be favorable for the growth of yeasts; the air probably contains large numbers of yeast cells.

A MOTHER-STARTER CAN.

To overcome the difficulty from yeast contamination some means are necessary to insure a pure mother-starter which is protected from

contact with the air after sterilizing. A can or receptacle which works very satisfactorily was devised by one of the authors (see text fig. 1). This starter-can consists of two tanks (5) and (11) connected by a block-tin collar (7)—brass can not be used, as it prevents growth of the culture. In this collar is fitted a tinned plug (8), fastened to a tinned-brass pipe (6). A brass collar (1) large enough to

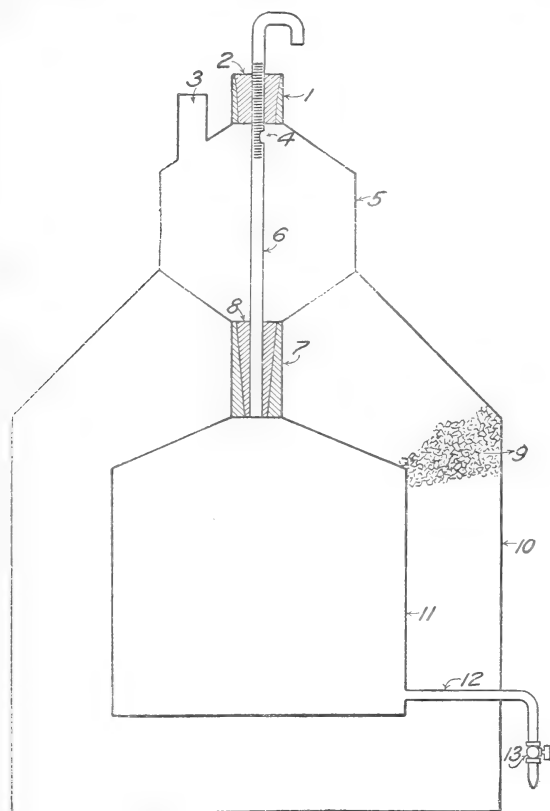


FIG. 1.—A mother-starter can for carrying pure cultures. Although it was designed especially to prevent cultures of *Bacillus bulgaricus* from yeast contamination in making Swiss cheese, it can be used for all whey cultures.

let the block-tin plug pass through is fastened at the top of the upper tank. Into this brass collar is fitted a brass plug (2), through which passes the tinned-brass pipe (6). Both the pipe and plug are threaded to permit the raising and lowering of the block-tin plug (8) between the two tanks without raising the brass plug in the top tank. When both plugs are in place there should be an opening in the pipe at the point (4) to permit the air to pass from the lower tank (11) to the upper tank (5) when the whey is passing from the upper tank to the lower tank. The end of the brass pipe (6) extending above the

upper tank should be plugged with cotton to prevent any outside contamination when the starter is drawn from the lower tank. The opening (3) in the top of the upper tank for filling is plugged with a cork into which is fitted a thermometer. The pipe (12) for draining off the starter is placed about one-half inch from the bottom of the lower tank. This always leaves enough starter for reinoculation. The lower can is insulated with a zinc jacket filled with ground cork (9).

PREPARING THE STARTER.

The starter can is first sterilized with steam or boiling water. The pure culture is then placed in the lower tank and the block-tin plug screwed down in place. The upper tank is then filled with boiling whey and the cork and thermometer inserted. The whey is allowed to stand in the upper tank until cooled to about 104° F. if *Bacillus bulgaricus* starter is used (in case the common lactic-acid type is used, the temperature should be 77° F.). The block-tin plug is then raised by means of the brass pipe to let the whey pass to the lower tank, where it is inoculated with the bacteria in the pure culture previously placed there. The block-tin plug is then replaced. The next day this starter is drawn off and the upper tank refilled with boiling whey, this operation being repeated from day to day. It is desirable to fill the upper chamber with the boiling whey in order that the chamber may be completely sterilized and all yeast cells destroyed. Milk can not be used in this apparatus, because the coagulated casein might clog the small pipe at the bottom of the lower chamber.

It would appear probable that the starter-can described above might be used for carrying mother starters for all dairy purposes, if whey is used. It could be made in any size to suit, and it might be used for carrying the entire starter. Where used for a mother starter in a Swiss-cheese factory, the upper chamber should hold almost a pint and the lower chamber 1½ pints. If it is used for carrying the entire starter, the upper chamber should hold 5 gallons and the lower chamber slightly more. For a Cheddar-cheese factory a can three times as large as for a Swiss-cheese factory might be needed. As already indicated, the mother starter for which the can is designed is a small starter carried under conditions to insure its remaining pure. The mother starter is added to the larger starter intended for the milk.

To prepare the starter where this mother-starter can is used the whey used should be first heated by setting in boiling water and holding for not less than 15 minutes at about 200° F. It should then be cooled to 100° F. and 2 to 5 per cent of the mother starter added. The whey with the rennet added should then be carried at 100° F. for 24 hours, when it should have more than 1 per cent of acid. If the mother starter becomes contaminated with yeast, which may happen if whey from some other factory or any other means than a pure culture is used to renew the starter, the can should be drained and filled with boiling water two or three times successively to kill the yeast present and another start made with the culture or the whey, as the case may be.

A growth of *Mycoderma*, which shows itself on the starter as a thin white film, is desirable for improving the efficiency of the starter.

This could be grown in the mother starter, and if the whey mother starter is drawn to the level of the tube in the starter-can it will carry enough of the *Mycoderma* to inoculate the starter.

SUMMARY AND CONCLUSIONS.

Many cultures of *Bacillus bulgaricus* obtained from different sources were used as starters in experiments for suppressing gas-forming bacteria in milk used for making Swiss cheese.

The ability to suppress undesirable fermentations was found to vary widely with different strains of *B. bulgaricus*. Several were able to prevent gas formation when the starter was less than 2 per cent of the total amount of milk used. Other cultures did not prove efficient with less than 4 per cent.

Ordinary lactic-acid cultures were not successful in preventing gas formation.

The application of the use of *B. bulgaricus* as a starter was tried in a cheese factory under commercial conditions. Good cheese was made at a season when it was not possible to make marketable Swiss cheese without the use of the cultures.

The results of these experiments indicate that the maker of Swiss cheese can control the fermentations with some cultures of *B. bulgaricus*; that a good quality of Swiss cheese can be made in winter as well as in summer; and that it is probably practicable to make cheese once a day instead of twice a day, as has been necessary in the past.

Methods are described for preparing and keeping cultures. A new type of starter-can for carrying starter is illustrated and described. This starter-can may be used for other dairy purposes with whey starters.

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